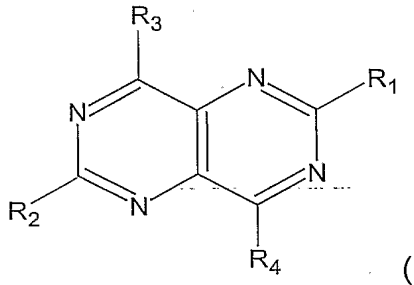
CLAIMS

1. Use of inhibitors of h-Prune cyclic nucleotide phosphodiesterase activity for the preparation of a medicament for prevention and treatment of tumour metastases characterised by an overexpression of h-PRUNE.

2. Use according to claim 1, wherein inhibitors of h-prune cyclic nucleotide phosphodiesterase activity have the general formula (I):



(I)

wherein R1 and R2, which are the same or different, can be selected from the group consisting of amino alcohol, amino alkyl, cholesterol;
wherein R3 and R4, which are the same or different, can be selected from the group consisting of eterocyclic aromatic or aromatic rings.

3. Use according to claim 2, wherein said eterocyclic aromatic rings can be selected from the group consisting of pyrazole, pyrrole, imidazole, pyridine, pyrimidine, morpholine.

4. Use according to any one of claims 2 and 3, wherein R1 and/or R2 are diethanolamine

5. Use according to any one of claims from 2 to 4, wherein R3 and/or R4 are pyrimidine.

6. Use according to any one of claims from 2 to 5, wherein said inhibitor is dipyridamole.

7. Use according to claim 1, wherein inhibitors of h-PRUNE cyclic nucleotide phosphodiesterase activity are selected from the group consisting of vinpocetine, 3-isobutyl-1-methylxanthine, IC261 and derivatives, structural analogues and isomers thereof.

8. Use according to the claim 1, wherein inhibitor of h-prune cyclic nucleotide phosphodiesterase activity is the peptide comprising the following amino acidic sequence:
NIIHGSOSVESAEKE (SEQ ID No 9).

9. Use according to the claim 1, wherein inhibitor of h-prune cyclic nucleotide phosphodiesterase activity is the peptide comprising the following amino acidic sequence: NIIHGSOSVESAEKE GGGYGRKKRRORRR (SEQ ID No 10); and characterised in that it is permeable.

10. Use according to any one of preceding claims, wherein tumours characterised by an overexpression of h-PRUNE are breast carcinoma, sarcoma, neuroblastoma, prostate tumour, pancreatic tumour, colonic tumour, rectal tumour, medulloblastoma, epitelioma, epatocarcinoma, cell T or cell B lymphomas, myeloma and melanoma, and pulmonary tumour.

11. Peptide comprising the following amino acidic sequence: NIIHGSOSVESAEKE (SEQ ID No 9).

12. Peptide comprising the following amino acidic sequence: NIIHGSOSVESAEKEGGGYGRKKRRQRRR (SEQ ID No 10) characterised in that it is permeable.

13. Screening method for h-PRUNE-inhibiting compounds, comprising the following phases:

a) selection of at least a phosphoesterase (PDE) inhibiting compound or derivative, structural analogue or isomer thereof;

b) administration of said at least one compound at concentration between 0,05 μM and 10 μM in a cell line overexpressing h-PRUNE;

c) quantitative analysis of the cyclic nucleotide phosphodiesterase activity of h-PRUNE and/or analysis of cellular motility versus concentration of said at least one compound and chemo-attractant and selection of compound able to inhibit said phosphodiesterase activity between the values from 0.01 to 1 $pmol/min^{-1}/ug^{-1}$ and/or inhibit said motility up to the attainment of the values between 200 and 1200 cells.

14. Method according to claim 13, wherein the cellular line is MDA-C100 435 prune #4.

15. Method according to any one of claims 13 and 14, wherein the quantitative analysis is carried out by hydrolysis tests of the c-AMP and/or c-GMP substrate.

16. Method according to claim 15, wherein the substrate is used at concentration between 0,008 μM and 1 μM.

17. Method for detection of h-PRUNE in a biological sample for metastases diagnosis of tumours characterised by an h-PRUNE overexpression by immunological assay, FISH analysis, Real-time PCR, *in situ* hybridization.

18. Method according to claim 17, comprising the following steps:
a) bring into contact said biological sample with at least one anti-h-PRUNE antibody;
b) detection of the antigen-antibody complex;
c) quantitative analysis of the antigen-antibody complex.

19. Method according to claim 18, wherein said biological sample is a tissue section or biological fluid.

20. Method according to any one of claims from 17 to 19, wherein said anti-h-PRUNE antibody is a monoclonal or polyclonal antibody.

21. Method according to any one of claims from 17 to 20, wherein said anti-h-PRUNE antibody is labelled with a radioisotope, fluorescent molecule or enzyme.

22. Method according to claim 18, wherein said detection and quantitative analysis of the antigen-antibody complex are performed by immunohistochemistry, immunoprecipitation, immunofluorescence, ELISA, immunoblotting analyses.

23. Method according to claim 17, wherein PCR Real time primers specific for h-PRUNE comprise the sequences:
5'-AGAGATCTTGGACAGGCAAACT-3' (SEQ ID No 1);
3'-CCATGTTGACACAGTCCAGGAT-5' (SEQ ID No 2);
or their complementary sequences.

24. Method according to claim 17, wherein the labelled probe for Real-time PCR or *in situ* hybridization comprise the oligonucleotidic sequence:
CTGCATGGAACCATC (SEQ ID No 3)
or its complementary sequence or the sequence wherein T is replaced by U.

25. Method according to claim 24, wherein said labelled probe for Real-time PCR is linear or circular one.

26. Method according to any one of claims 24 and 25, wherein said probe is labelled with at least one radioisotope and/or fluorochrome.

27. Method according to any one of claims from 24 to 26, wherein said probe is labelled with at least a fluorochrome at 5' and/or 3'.

28. Method according to any one of claims from 24 to 25, wherein said fluorochrome is 6-carboxifluorescein.

29. Diagnostic kit for the detection of h-PRUNE in a biological sample for metastases diagnosis of tumours characterised by an h-PRUNE over-expression comprising at least one anti-h-PRUNE antibody, or a pair of

primers specific for h-PRUNE or labelled oligonucleotidic probe specific for h-PRUNE.

30. Diagnostic kit according to claim 29, wherein the tumours characterised by an h-PRUNE overexpression are breast carcinoma, sarcoma, neuroblastoma, melanoma.

31. Diagnostic kit according to any one of claims 29 and 30, wherein said anti-h-PRUNE antibody is monoclonal or polyclonal antibody.

32. Diagnostic kit according to claim 31, wherein said anti-h-PRUNE antibody is labelled with a radioisotope, fluorescent molecule or enzyme.

33. Diagnostic kit according to claim 29, wherein said pair of primers specific for h-PRUNE comprises the sequences:
5'-AGAGATCTTGGACAGGCAAACT-3' (SEQ ID No 1);
3'-CCATGTTGACACAGTCCAGGAT-5' (SEQ ID No 2);
or their complementary sequences.

34. Diagnostic kit according to claim 29, wherein said labelled oligonucleotidic probe for Real-time PCR or *in situ* hybridization comprises the oligonucleotidic sequence:
CTGCATGGAACCATC (SEQ ID No 3)
or its complementary sequence or the sequence wherein T is replaced by U.

35. Diagnostic kit according to claim 34, wherein said labelled oligonucleotidic probe for Real-time PCR is linear or circular one.

36. Diagnostic kit according to any one of claims 34 and 35, wherein said oligonucleotidic probe is labelled with at least one radioisotope and/or fluorochrome.

37. Diagnostic kit according to any one of claims from 34 to 36, wherein said probe is labelled with at least one fluorochrome at 5' and/or 3'.

38. Diagnostic kit according to claim 37, wherein the fluorochrome is 6-carboxifluorescein.

39. Monoclonal murine antibody able to recognise and bind selectively the h-PRUNE recombinant protein, characterised in that it belongs to the IgM immunoglobulin class and is produced by 4G3/4 clone (deposited at the CBA in Genoa on 10/12/2004)

40. Polyclonal antibody from rabbit for h-PRUNE characterised in that it recognises and binds selectively the peptide used for the immunisation comprising the amino acidic sequence:

NH_2-Ala-Leu-Glu-Glu-Ala-Val-Ala-Glu-Val-Leu-Asp-His-Arg-Pro-Ile-Glu-Pro-Lys-COOH (SEQ ID No 4)
or parts thereof.

41. Specific primers for hPRUNE amplification through Real-time PCR comprising at least one of the oligonucleotidic sequences:
5'-AGAGATCTTGGACAGGCAAACT-3'(SEQ ID No 1);
3'-CCATGTTGACACAGTCCAGGAT-5'; (SEQ ID No 2);
or their complementary sequences.

42. Oligonucleotidic probe specific for h-PRUNE for Real-time PCR or *in situ* hybridization comprising the sequence
CTGCATGGAACCATC (SEQ ID No 3);
or its complementary sequence or the sequence wherein T is replaced by U.

43. Oligonucleotidic probe according to claim 42, wherein said probe is linear or circular one.

44. Oligonucleotidic probe according to any one of claims 42 and 43, wherein said probe is labelled with at least one radioisotope and/or fluorochrome.

45. Oligonucleotidic probe according to any one of claims from 42 to 44, wherein said probe is labelled with at least one fluorochrome at 5' and/or 3'.

46. Oligonucleotidic probe according to claim 45, wherein the fluorochrome is 6-carboxifluorescein.